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=> s two (w) hybrid and (penicillin? or cephalosporin?)
L1            9 TWO (W) HYBRID AND (PENICILLIN? OR CEPHALOSPORIN?)

=> dup rem l1
PROCESSING COMPLETED FOR L1
L2            6 DUP REM L1 (3 DUPLICATES REMOVED)

=> d 1-6 ti

L2   ANSWER 1 OF 6  CAPLUS  COPYRIGHT 2004 ACS on STN
TI   Interaction trap assays using selectable markers to screen large libraries
     for protein-protein and protein-nucleic acid interactions

L2   ANSWER 2 OF 6     MEDLINE on STN                    DUPLICATE 1
TI   Transcription of the gene mediating methicillin resistance in
     Staphylococcus aureus (mecA) is corepressed but not coinduced by cognate
     mecA and beta-lactamase regulators.

L2   ANSWER 3 OF 6  CAPLUS  COPYRIGHT 2004 ACS on STN
TI   A generic complementation assay for protein evolution and proteomics

L2   ANSWER 4 OF 6     MEDLINE on STN                    DUPLICATE 2
TI   The fungal CPCR1 protein, which binds specifically to beta-lactam
     biosynthesis genes, is related to human regulatory factor X transcription
     factors.

L2   ANSWER 5 OF 6     MEDLINE on STN                    DUPLICATE 3
TI   A fast method to predict protein interaction sites from sequences.

L2   ANSWER 6 OF 6     MEDLINE on STN
TI   Construction of a cassette for cloning and analysis of replicons.


=> d 1-6 bib ab

L2   ANSWER 1 OF 6  CAPLUS  COPYRIGHT 2004 ACS on STN
AN   2001:851433  CAPLUS
DN   136:1569
TI   Interaction trap assays using selectable markers to screen large libraries
     for protein-protein and protein-nucleic acid interactions
IN   Joung, J. Keith; Miller, Jeffrey; Pabo, Carl O.
PA   Massachusetts Institute of Technology, USA
SO   PCT Int. Appl., 196 pp.
     CODEN: PIXXD2
DT   Patent
LA   English
FAN.CNT 2
     PATENT NO.         KIND  DATE          APPLICATION NO.  DATE
     ---------------    ----  --------      ---------------  --------
PI   WO 2001088197       A2   20011122      WO 2001-US15718  20010516
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WO 2001088197 A3 20031231
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
PRAI US 2000-204509P P 20000516
AB The present invention provides methods and compns. for interaction trap assays for detecting protein-protein, protein-DNA, or protein-RNA interactions using prokaryotic or microbial eukaryotic hosts. The methods and compns. of the invention may also be used to identify agents which may agonize or antagonize a protein-protein, protein-DNA, or protein-RNA interaction. In certain embodiments, the interaction trap system of the invention is useful for screening libraries with greater than 107 members. In other embodiments, the interaction trap system of the invention is used in conjunction with flow cytometry. The invention further provides a means for simultaneously screening a target protein or nucleic acid sequence for the ability to interact with two or more test proteins or nucleic acids. In one form, the screening involves the use of a selectable marker allowing screening of large nos. of cells without the need to scan for a colorimetric marker. In a second form, screening of a colorimetric marker is by flow cytometry. Screening of a library of 108 members in Escherichia coli for C2H2 zinc finger variants is demonstrated.

L2 ANSWER 2 OF 6 MEDLINE on STN DUPLICATE 1
AN 2001645967 MEDLINE
DN PubMed ID: 11698375
TI Transcription of the gene mediating methicillin resistance in Staphylococcus aureus (mecA) is corepressed but not coinduced by cognate mecA and beta-lactamase regulators.
AU McKinney T K; Sharma V K; Craig W A; Archer G L
CS Department of Medicine, Virginia Commonwealth University, Medical College of Virginia Campus, 1101 E. Marshall St., Richmond, VA 23298-0049, USA.
NC R37 AI 35705 (NIAID)
SO Journal of bacteriology, (2001 Dec) 183 (23) 6862-8.
Journal code: 2985120R. ISSN: 0021-9193.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200112
ED Entered STN: 20011108
Last Updated on STN: 20020123
Entered Medline: 20011207
AB Resistance to beta-lactam antibiotics in staphylococci is mediated by mecA and blaZ, genes encoding a **penicillin**-binding protein (PBP2a) with low beta-lactam affinity and beta-lactamase, respectively. The mec and bla regulators, mecR1-mecI and blaR1-blaI, respectively, encode inducer-repressors with sufficient amino acid homology to suggest that they could coregulate PBP2a production. In order to test this hypothesis, plasmids containing mec and bla regulatory sequences were introduced into Staphylococcus aureus containing a chromosomal mecA-lacZ transcriptional fusion. Corepression was confirmed by demonstrating a gene dosage-dependent reduction in beta-galactosidase activity by either MecI or BlaI and additive repression when both were present. Both MecI-MecI and BlaI-BlaI homodimer and MecI-BlaI heterodimer interactions were demonstrated in the yeast **two-hybrid** assay, and purified MecI and BlaI protected the same mec promoter-operator sequences. However, MecI was approximately threefold more effective at mecA-lacZ

transcriptional repression than was BlaI. While MecI and BlaI displayed similar activity as repressors of mecA transcription, there was a marked difference between MecR1 and BlaR1 in the rate and specificity of induction. Induction through BlaR1 by a beta-lactam was 10-fold greater than through MecR1 at 60 min and was 81% of maximal by 2 h, while induction through MecR1 never exceeded 20% of maximal. Furthermore, complementation studies showed that MecI- or BlaI-mediated mecA transcriptional repression could be relieved by induction through homologous but not heterologous sensor-inducer proteins, demonstrating the repressor specificity of induction.

L2 ANSWER 3 OF 6 CAPLUS COPYRIGHT 2004 ACS on STN
AN 2001:637015 CAPLUS
TI A generic complementation assay for protein evolution and proteomics
AU Krane, Sonja; Bleczinski, Colleen F.; Baker, Kathleen; Lin, Hening; Salazar-Jimenez, Gilda J.; Sengupta, Debleena; Cornish, Virginia W.
CS Department of Chemistry, Columbia University, New York, NY, 10027, USA
SO Abstracts of Papers, 222nd ACS National Meeting, Chicago, IL, United States, August 26-30, 2001 (2001), BIOL-004 Publisher: American Chemical Society, Washington, D. C.
CODEN: 69BUZP
DT Conference; Meeting Abstract
LA English
AB Reaction-independent assays for enzymic activity hold tremendous promise for protein evolution and proteomics. The difficulty is to design a method that is high-throughput and that can readily be adapted to a variety of different chemical reactions. Here we describe an approach, based on the yeast **two-hybrid** assay, where we make transcription of a reporter gene dependent on enzymic turnover. Specifically, dimerization of the DNA-binding and activation domains of a transcriptional activator is made dependent on a dimeric ligand, or chemical inducer of dimerization (CID), and then the bond between the two ligands is replaced with the chemical of interest. Thus enzyme-catalyzed cleavage or formation of the bond between the two ligands controls reconstitution of the transcriptional activator and transcription of an engineered reporter gene. As a proof of principle, we have used a well-studied enzymic reaction, cephem hydrolysis by a **cephalosporinase**, to demonstrate this strategy. First we synthesized a dexamethasone-methotrexate (Dex-Mtx) CID with a cleavable cephem linker (Dex-cephem-Mtx) and showed that this mol. retained the ability to activate transcription in the yeast **two-hybrid** assay. Then we introduced the **cephalosporinase** enzyme into the yeast cells and developed conditions where we can detect enzyme-catalyzed cleavage of the cephem linker as disruption of the transcriptional read-out. Finally, in a mock screen, we have distinguished active and inactive **cephalosporinase** variants based on their effect on the levels of transcription of a lacZ reporter gene.

L2 ANSWER 4 OF 6 MEDLINE on STN DUPLICATE 2
AN 2000200423 MEDLINE
DN 20200423 PubMed ID: 10734077
TI The fungal CPCR1 protein, which binds specifically to beta-lactam biosynthesis genes, is related to human regulatory factor X transcription factors.
AU Schmitt E K; Kuck U
CS Lehrstuhl fur Allgemeine und Molekulare Botanik, Ruhr-Universitat Bochum, D-44780 Bochum, Germany.
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (2000 Mar 31) 275 (13) 9348-57.
Journal code: 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals

OS GENBANK-AJ132014; GENBANK-AJ243296
EM 200005
ED Entered STN: 20000512
Last Updated on STN: 20000512
Entered Medline: 20000504
AB Here we report the isolation and characterization of a novel transcription factor from the **cephalosporin** C-producing fungus Acremonium chrysogenum. We have identified a protein binding site in the promoter of the beta-lactam biosynthesis gene pcbC, located 418 nucleotides upstream of the translational start. Using the yeast one-hybrid system, we succeeded in isolating a cDNA clone encoding a polypeptide, which binds specifically to the pcbC promoter. The polypeptid shows significant sequence homology to human transcription factors of the regulatory factor X (RFX) family and was designated CPCR1. A high degree of CPCR1 binding specificity was observed in in vivo and in vitro experiments using mutated versions of the DNA binding site. The A. chrysogenum RFX protein CPCR1 recognizes an imperfect palindrome, which resembles binding sites of human RFX transcription factors. One- and **two-hybrid** experiments with truncated versions of CPCR1 showed that the protein forms a DNA binding homodimer. Nondenaturing electrophoresis revealed that the CPCR1 protein exists in vitro solely in a multimeric, probably dimeric, state. Finally, we isolated a homologue of the cpcR1 gene from the **penicillin**-producing fungus Penicillium chrysogenum and determined about 60% identical amino acid residues in the DNA binding domain of both fungal RFX proteins, which show an overall amino acid sequence identity of 29%.

L2 ANSWER 5 OF 6 MEDLINE on STN DUPLICATE 3
AN 2001009082 MEDLINE
DN PubMed ID: 10993732
TI A fast method to predict protein interaction sites from sequences.
AU Gallet X; Charloteaux B; Thomas A; Brasseur R
CS Centre de Biophysique Moleculaire Numerique, Faculte Agronomique, Gembloux, 5030, Belgium.. brasseur.r@fsagx.ac.be
SO Journal of molecular biology, (2000 Sep 29) 302 (4) 917-26.
Journal code: 2985088R. ISSN: 0022-2836.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200010
ED Entered STN: 20010322
Last Updated on STN: 20010322
Entered Medline: 20001025
AB A simple method for predicting residues involved in protein interaction sites is proposed. In the absence of any structural report, the procedure identifies linear stretches of sequences as "receptor-binding domains" (RBDs) by analysing hydrophobicity distribution. The sequences of two databases of non-homologous interaction sites eliciting various biological activities were tested; 59-80 % were detected as RBDs. A statistical analysis of amino acid frequencies was carried out in known interaction sites and in predicted RBDs. RBDs were predicted from the 80,000 sequences of the Swissprot database. In both cases, arginine is the most frequently occurring residue. The RBD procedure can also detect residues involved in specific interaction sites such as the DNA-binding (95 % detected) and Ca-binding domains (83 % detected). We report two recent analyses; from the prediction of RBDs in sequences to the experimental demonstration of the functional activities. The examples concern a retroviral Gag protein and a **penicillin**-binding protein. We support that this method is a quick way to predict protein interaction sites from sequences and is helpful for guiding experiments such as site-specific mutageneses, **two-hybrid** systems or the synthesis of inhibitors.

L2 ANSWER 6 OF 6 MEDLINE on STN
AN 2000441227 MEDLINE
DN 20430401 PubMed ID: 10974702
TI Construction of a cassette for cloning and analysis of replicons.
AU Alonso G; Campos J; Bruzual I; Rodriquez Lemoine V
CS Laboratorio de Biologia de Plasmidos, Facultad de Ciencias, Universidad Central de Venezuela.. galonso@reacciun.ve
SO ACTA CIENTIFICA VENEZOLANA, (2000) 51 (1) 4-9.
Journal code: 0070154. ISSN: 0001-5504.
CY Venezuela
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200009
ED Entered STN: 20000928
Last Updated on STN: 20000928
Entered Medline: 20000919
AB The aim of this work was the construction of a cassette, i.e., a non-replicative molecule formed by linkage of an antibiotic resistance gene and a multiple cloning site. This cassette would allow the cloning and analysis of a wide range of replicons. The aac(6')-1c amikacin gene was isolated and ligated to the multiple cloning site of the pUC18 vector. This construction was HindIII digested and cloned in the HindIII site of the vector. The resulting pHJ13 clone conferred to the recipient cells the ability to grow in presence of amikacin (cassette marker) and ampicillin (vector gene). By restriction analysis, the cassette orientation was established. Cassette versatility is provided by the presence of the unaltered multiple cloning site segment, and also because it allows sequencing of any replication origin inserted. Cassette functionality was demonstrated by ligation to a replicative region of H plasmid pHH1457. Presence of the ori region from pHH1457 and the aac(6')-1c gene was confirmed in E. coli transformed clones. The incompatibility properties of the pHH1457 and its capability to replicate in a Poll defective strain were preserved in the pHJII14 construct. Currently, the amikacin cassette is being used in the characterization of H Complex plasmids.

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L Number	Hits	Search Text	DB	Time stamp
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2	29	(two adj1 hybrid) same (penicillin or cephalosporin)	USPAT; US-PGPUB	2004/02/26 15:46
3	20	mitchnick$.in.	USPAT; US-PGPUB	2004/02/26 15:47
4	20	galarneau$.in.	USPAT; US-PGPUB	2004/02/26 15:47

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